132 Non-respiratory swabs have little role in the detection of methicillin resistant *Staphylococcus aureus* in cystic fibrosis

A. Horsley[1], E. Carassale[2], M. Cullen[2], B. Isalska[2], R. Bright-Thomas[1], K. Webb[1], A. Jones[1]. [1]*Manchester Adult CF Centre, Manchester, United Kingdom;* [2]*University Hospitals of South Manchester, Dept of Microbiology, Manchester, United Kingdom*

Background: Chronic methicillin resistant *Staphylococcus aureus* (MRSA) infection in CF is associated with faster FEV1 decline and greater mortality. Screening for infection involves swabs from both upper respiratory (nose, throat) and peripheral sites (groin, axilla), as well as sputum culture. There are no data to indicate which is superior.
Methods: Microbiology results of pts attending our adult tertiary care centre who have had any positive MRSA result between 01/01/09 and 01/08/2010 were reviewed. Patients were considered continuously infected between their earliest and last MRSA culture.
Results: 17pts (5%) had at least 1 MRSA isolate: 6 known chronic MRSA; 3 intermittent; 6 detected on routine outpatient sputum culture; 2 detected on ward screening.
307 microbiology samples were taken during periods of MRSA positivity: 180 (59%) were swabs (throat = 71, nose = 35, groin = 59 and axilla = 15) and 127 sputa. Of these, 70 (55%) sputa were MRSA positive. Nose swabs grew MRSA in 24%, throat 23%, groin 19% and axilla 0% cases.
28 sets of concurrent samples from 12 pts were discordant. Nose swabs were positive in 17/28 (61%) of these, throat swabs in 8/14 (57%). Peripheral swabs grew MRSA in 5/27 (19%), all also positive in nose +/− sputum. Concurrent sputa were MRSA positive in 15/23 (65%).
Discussion: Upper respiratory tract swabs constitute an important tool for MRSA surveillance. Little additional information however is derived from non-respiratory swabs, and their routine use could be discontinued in units with low MRSA prevalence. In those who expectorate easily, sputum is the most effective method of detecting MRSA. There is a considerable false negative rate for all sample methods.

133* Characteristics and susceptibilities of methicillin resistant *Staphylococcus aureus* in paediatric cystic fibrosis patients in the USA

M.S. Muhlebach[1], E. Champion[1], E. Popowitch[2], M.B. Miller[2], L. Saiman[3], STAR-CF sites. [1]*University North Carolina (UNC), Pediatrics, Chapel Hill, United States;* [2]*UNC, Microbiology, Chapel Hill, United States;* [3]*University of Columbia, New York, United States*

Background: Prevalence of methicillin resistant *Staphylococcus aureus* (MRSA) in CF has increased to ~22% in the US. This parallels rises of healthcare associated MRSA (HA-MRSA) and MRSA in the community (CA), especially strain USA300 (SCC*mec*IV PVL+). This study examines the prevalence of HA- vs. CA-MRSA based on molecular typing; antimicrobial susceptibility testing (AST) and patient characteristics at seven US CF centres.
Methods: MRSA isolates are collected randomly from paediatric CF patients and are typed by SCC*mec* and *pvl* status. AST is done using VITEK2 GP67 card and Etest for fusidic acid, fosfomycin and mupirocin according to US and European Microbiological Guidelines. Clinical data are obtained from the US CFF Patient Registry.
Results: Of 299 MRSA isolates, 69% (range 63–83%) are SCC*mec*II (HA-MRSA). The remaining isolates are SCC*mec*IV of which 57% are *pvl* positive. Resistance (R) is ~90% for erythromycin for all molecular types; Antibiotics with least R were TMP-SMX (3–10%), tetracyclines (4–8%) and fusidic acid (0–5%). Clindamycin R is high in HA and low in SCC*mec*IV strains. No isolates are resistant to vancomycin and 1/299 each resistant to linezolid or tigecyclcine. Mupirocin R was 8% in CA- and 0 in HA-MRSA. Presently clinical information is available on 193 patients showing a mean age of 11.5 ± 4.7 years and 71% with chronic *P. aeruginosa*.
Conclusion: In the US, about 1/3 of MRSA strains among paediatric CF patients are SCC*mec*IV of which half are *pvl* positive. Despite ample use resistance to TMP-SMX is low but fluoroquinolone R high in HA-MRSA. Although not being approved in the US sporadic resistance to fosfomycin and fusidic acid occurs. Link to demographic data for all patients is ongoing.

134* Molecular characterization of methicillin-resistant *Staphylococcus aureus* (MRSA) strains responsible for the first infection in cystic fibrosis patients

P. Cocchi[1], G. Taccetti[2], E. Fiscarelli[3], L. Cariani[4], C. Braggion[2], M. de Martino[1], S. Campana[2]. [1]*University of Florence, Department of Sciences for Woman and Child's Health, Florence, Italy;* [2]*Cystic Fibrosis Center, Anna Meyer Children's University Hospital, Department of Pediatric Medicine, Florence, Italy;* [3]*Bambino Gesù Children's Hospital – IRCCS, Rome, Italy;* [4]*Cystic Fibrosis Center Microbiology Laboratory, Milan, Italy*

Background: Recent studies have shown that MRSA is associated with of worse survival in CF patients. The role of a different genetic background in community-acquired (CA-MRSA) and hospital-acquired (HA-MRSA) strains in this process remains unclear. There is no evidence of the preponderant role that particular clones might have in worsening CF patients conditions.
Aim: The aim of this study was to analyze MRSA strains responsible for first infections of CF airways, and determine their genetic background.
Materials and Methods: 42 strains infecting for the first time the airways of 42 CF patients were collected and analyzed determining SCC*mec* type and sequence type as previously described (Oliveira and de Lancastre, 2002 and Enright *et al*, 2000).
Results: Twenty (47.6%) out of 42 strains belong to CA-MRSA, the remaining 22 (52.4%) to HA-MRSA. MLST analysis, performed on 24 out of 42 strains, showed the presence of already described clones, including ST8-MRSA-IV, ST228-MRSA-I or Southern Germany clone, and ST45-MRSA-IV or Berlin clone.
Conclusions: Our study suggests that the prevalence of CA-MRSA and HA-MRSA is comparable and demonstrates that known epidemic clones are responsible for first infection. Further studies are needed to clarify the role of these clones on clinical status of CF patients.
This work was supported by Italian Cystic Fibrosis Research Foundation (grant FFC#11 2009) with the contribution of Angelini.

136* Overlapping genes as molecular markers for identification of bacteria: the use of *marC–hisH* for the identification of *Burkholderia* and *Ralstonia*

E. Perrin[1], M.C. Papaleo[1], I. Maida[1], M. Fondi[1], M. Vaneechoutte[2], R. Fani[1]. [1]*University of Florence, Department of Evolutionary Biology 'Leo Pardi', Florence, Italy;* [2]*University of Gent, Lab. Bacteriology Research (LBR), Gent, Belgium*

CF patients are susceptible to respiratory tract infection with a variety of bacterial species including species belonging to the genera *Burkholderia* and *Ralstonia*. The role of *Burkholderia* in contributing to pulmonary disease in CF is well known, whereas little is known about the relevance of *Ralstonia*. This is also due to the difficult identification of *Ralstonia* species, which are frequently misidentified as other closely related species.
Thus, the aim of this work was to set up a diagnostic method to identify *Ralstonia* and *Burkholderia* strains by applying a new molecular strategy based on the use of nested overlapping genes. To this purpose, we focused on genes of the histidine (*his*) operon, because the analysis of 1098 genomes revealed the insertion of *marC*, a gene unrelated to *his* biosynthesis, within *his* operon, whereby *marC* partially overlaps with *hisH* **only** in **all** strains belonging to these two genera. Hence, three forward PCR primers (one specific for *Burkholderia*, one for *Ralstonia* and one for both genera) targeting *marC* and one reverse primer targeting *hisH* were designed. The three primer sets were tested on 35 *Burkholderia* and 22 *Ralstonia* strains and the expected amplicons were obtained for all strains analyzed. Sequencing of amplicons confirmed the correct amplification. All other Gram-negative nonfermenting species tested remained negative, with the interesting exception of *Cupriavidus* (*Wautersia*) *respiraculi*, previously regarded as belonging to the genus *Ralstonia,* which was positive with the general primer but not with the two species specific primers.